abcam

ab227843

Anti-RIP antibody [EPR4689-100] - BSA and Azide free

Recombinant | RabMAb | KO Validated

Key facts

Isotype	IgG
Host species	Rabbit
Storage buffer	pH: 7.2 - 7.4 Constituents: PBS
Form	Liquid
Clonality	Monoclonal
Immunogen	The exact immunogen used to generate this antibody is proprietary information.
Carrier free	Yes
Clone number	EPR4689-100
Purification technique	Affinity purification Protein A
Concentration	1.014 mg/mL The concentration of this product may be batch-dependent Batch concentration finder →

Reactivity data

ICC/IF

Not recommended

Species	Human
Dilution info	-

Notes	-

IP

Not recommended

Species	Human
Dilution info	-
Notes	-

WB

Tested

Species	Human
Dilution info	-
Notes	-

IHC-P

Not recommended

Species	Human
Dilution info	-
Notes	-

Flow Cyt (Intra)

Tested

Species	Human
Dilution info	-
Notes	ab199376 - Rabbit monoclonal IgG, is suitable for use as an isotype control with this antibody.

Target data

See full target information RIPK1

Function	Serine-threonine kinase which is a key regulator of TNF-mediated apoptosis, necroptosis and inflammatory pathways (PubMed:17703191, PubMed:24144979, PubMed:31827280, PubMed:31827281, PubMed:32657447, PubMed:35831301). Exhibits kinase activity-dependent functions that regulate cell death and kinase-independent scaffold functions regulating inflammatory signaling and cell survival (PubMed:11101870, PubMed:19524512, PubMed:19524513, PubMed:29440439, PubMed:30988283). Has kinase-independent scaffold functions: upon binding of TNF to TNFR1, RIPK1 is recruited to the TNF-R1 signaling complex (TNF-RSC also known as complex I) where it acts as a scaffold protein promoting cell survival, in part, by activating the canonical NF-kappa-B pathway (By similarity). Kinase activity is essential to regulate necroptosis and apoptosis, two parallel forms of cell death: upon activation of its protein kinase activity, regulates assembly of two death-inducing complexes, namely complex IIa (RIPK1-FADD-CASP8), which drives apoptosis, and the complex IIb (RIPK1-RIPK3-MLKL), which drives necroptosis (By similarity). RIPK1 is required to limit CASP8-dependent TNFR1-induced apoptosis (By similarity). In normal conditions, RIPK1 acts as an inhibitor of RIPK3-dependent necroptosis, a process mediated by RIPK3 component of complex IIb, which catalyzes phosphorylation of MLKL upon induction by ZBP1 (PubMed:19524512, PubMed:19524513, PubMed:29440439, PubMed:30988283). Inhibits RIPK3-mediated necroptosis via FADD-mediated recruitment of CASP8, which cleaves RIPK1 and limits TNF-induced necroptosis (PubMed:19524512, PubMed:19524513, PubMed:29440439, PubMed:30988283). Required to inhibit apoptosis and necroptosis during embryonic development: acts by preventing the interaction of TRADD with FADD thereby limiting aberrant activation of CASP8 (By similarity). In addition to apoptosis and necroptosis, also involved in inflammatory response by promoting transcriptional production of pro-inflammatory cytokines, such as interleukin-6 (IL6) (PubMed:31827280, PubMed:31827281). Phosphorylates RIPK3: RIPK1 and RIPK3 undergo reciprocal auto- and trans-phosphorylation (PubMed:19524513). Phosphorylates DAB2IP at 'Ser-728' in a TNF-dependent manner, and thereby activates the MAP3K5-JNK apoptotic cascade (PubMed:15310755, PubMed:17389591). Required for ZBP1-induced NF-kappa-B activation in response to DNA damage (By similarity).

Storage

Shipped at conditions	Blue Ice
Appropriate short-term storage conditions	+4°C
Appropriate long-term storage conditions	+4°C
Storage information	Do Not Freeze

Notes

ab227843 is the carrier-free version of ab178420.

Species reactivity
Mouse, Rat: We have preliminary internal testing data to indicate this antibody may not react with these species. Please contact us for more information.

Conjugation ready
Our carrier-free antibodies are typically supplied in a PBS-only formulation, purified and free of BSA, sodium azide and glycerol. This conjugation-ready format is designed for use with fluorochromes, metal isotopes, oligonucleotides, and enzymes, which makes them ideal for antibody labelling, functional and cell-based assays, flow-based assays (e.g. mass cytometry) and Multiplex Imaging applications.

Use our conjugation kits for antibody conjugates that are ready-to-use in as little as 20 minutes with 1 minute hands-on-time and 100% antibody recovery: available for fluorescent dyes, HRP, biotin and gold.

Compatibility
This product is compatible with the Maxpar® Antibody Labeling Kit from Fluidigm, without the need for antibody preparation. Maxpar® is a trademark of Fluidigm Canada Inc.

Supplementary info

This supplementary information is collated from multiple sources and compiled automatically.

Activity summary	RIP also known as Receptor-Interacting Protein or RIPK1 is a serine/threonine-protein kinase with a mass of approximately 74 kDa. It plays an important role in cell death and survival signaling pathways. RIP is expressed ubiquitously across various tissues indicating its importance in many cellular functions. The protein contains a kinase domain an intermediate domain for protein-protein interactions and a death domain which facilitates its involvement in apoptotic signaling processes.
Biological function summary	Receptor-Interacting Protein Kinase 1 (RIPK1) participates in regulating both necroptosis and apoptosis distinguishing itself as an important mediator in cell death mechanisms. As part of the necrosome complex which includes RIPK3 and MLKL RIPK1 functions in necroptosis—a programmed form of necrosis. This characteristic involvement shows its dual role in maintaining cell fate decisions making it an integral part of immune response and inflammation control.
Pathways	RIPK1 strongly associates with the TNF signaling pathway and NF-kB pathway. Its interaction with TNF receptor 1 (TNFR1) and consequent involvement with TRADD and TRAF2 mediates the signal transduction necessary for the activation of NF-kB leading to transcription of genes involved in survival and inflammation. This connection illustrates its capability to switch between promoting cell survival through NF-kB and facilitating cell death via necroptosis or apoptosis depending on cellular context and cues.
Associated diseases and disorders	RIPK1 plays a significant role in conditions such as inflammatory diseases and neurodegenerative disorders. Its overactivation results in excessive cell death implicated in inflammatory conditions; necrostatin a necroptosis inhibitor targets RIPK1 to potentially mitigate this damage. Furthermore RIPK1's dysregulation links to Alzheimer's disease where it can interact with components like RIPK3 to exacerbate neurodegenerative processes. This relationship underlines the potential of targeting RIPK1 therapeutically to manage inflammation and neurodegeneration.

Product promise

Tested

We have tested this species and application combination and it works. It is covered by our product promise.

Expected

We have not tested this specific species and application combination in-house, but expect it will work. It is covered by our product promise.

Predicted

This species and application combination has not been tested, but we predict it will work based on strong homology. However, this combination is not covered by our product promise.

Not recommended

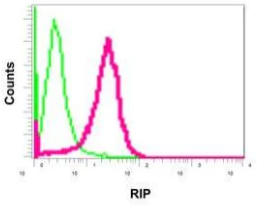
We do not recommend this combination. It is not covered by our product promise.

We are dedicated to supporting your work with high quality reagents and we are here for you every step of the way should you need us.

In the unlikely event of one of our products not working as expected, you are covered by our product promise.

Full details and terms and conditions can be found here:
Terms & Conditions.

4 product images

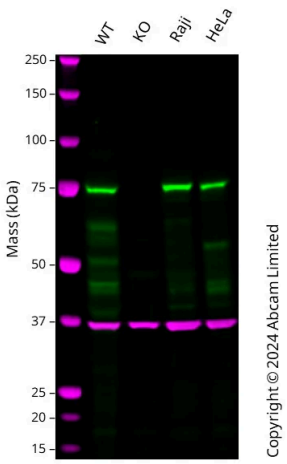


Flow Cytometry (Intracellular) - Anti-RIP antibody [EPR4689-100] - BSA and Azide free (ab227843)

RIP Flow Cytometry (Intracellular) staining using rabbit Anti-RIP antibody

This Flow Cyt data was generated using the same anti-RIP antibody clone, EPR4689-100, in a different buffer formulation (cat ab178420).

Intracellular flow cytometric analysis of permeabilized HeLa cells labeling RIP with ab178420 at 1/10 dilution (red) compared with a rabbit IgG negative control (green).



Western blot - Anti-RIP antibody [EPR4689-100] - BSA and Azide free (ab227843)

RIP Western blot staining using rabbit Anti-RIP antibody

This data was developed using the same antibody clone in a different buffer formulation (ab178420).

Western blot: Anti-RIPK1 antibody [EPR4689-100] (ab178420) staining at 1/1000 dilution, shown in green; Mouse anti-GAPDH antibody [6C5] (ab8245) loading control staining at 1/20000 dilution, shown in magenta. In Western blot, ab178420 was shown to bind specifically to RIPK1. A band was observed at 75 kDa in wild-type THP-1 cell lysates with no signal observed at this size in RIPK1 knockout cell line. To generate this image, wild-type and RIPK1 knockout THP-1 cell lysates were analysed. First, samples were run on an SDS-PAGE gel then transferred onto a nitrocellulose membrane. Membranes were blocked in 3 % milk in TBS-0.1 % Tween® 20 (TBS-T) before incubation with primary antibodies overnight at 4 °C. Blots were washed four times in TBS-T, incubated with secondary antibodies for 1 h at room temperature, washed again four times then imaged. Secondary antibodies used were Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution.

All lanes:
Western blot - Anti-RIP antibody [EPR4689-100] (ab178420) at 1/1000 dilution

Lane 1:
Wild-type THP-1 cell lysate at 20 µg

Lane 2:
RIPK1 knockout THP-1 cell lysate at 20 µg

Lane 3:
Raji cell lysate at 20 µg

Lane 4:
HeLa cell lysate at 20 µg

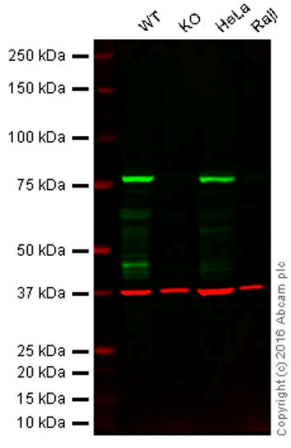
Secondary

All lanes:
Goat anti-Rabbit IgG H&L 800CW and Goat anti-Mouse IgG H&L 680RD at 1/20000 dilution

Performed under reducing conditions.

Predicted band size: 75 kDa

Observed band size: 75 kDa



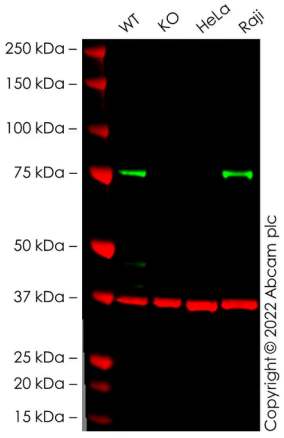
Western blot - Anti-RIP antibody [EPR4689-100] - BSA and Azide free (ab227843)

RIP Western blot staining using rabbit Anti-RIP antibody

This WB data was generated using the same anti-RIP antibody clone, EPR4689-100, in a different buffer formulation (cat# ab178420).
Lane 1: Wild-type HAP1 cell lysate (20 µg)
Lane 2: RIP knockout HAP1 cell lysate (20 µg)
Lane 3: HeLa cell lysate (20 µg)
Lane 4: Raji cell lysate (20 µg)
Lanes 1 to 4: Merged signal (red and green). Green - ab178420 observed at 78 kDa. Red - loading control, ab8245, observed at 37 kDa.

ab178420 was shown to specifically react with RIP when RIP knockout samples were used. Wild-type and RIP knockout samples were subjected to SDS-PAGE. ab178420 and ab8245 (loading control to GAPDH) were both diluted 1/1000 and 1/10000 respectively and incubated overnight at 4°C. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) ab216773 and Goat anti-Mouse IgG H&L (IRDye® 680RD) ab216776 secondary antibodies at 1/10000 dilution for 1 h at room temperature before imaging.
All lanes:
Western blot - Anti-RIP antibody [EPR4689-100] (ab178420)

Predicted band size: 75 kDa



Western blot - Anti-RIP antibody [EPR4689-100] - BSA and Azide free (ab227843)

All lanes:
Western blot - Anti-RIP antibody [EPR4689-100] (ab178420) at 1/1000 dilution

Lanes 1 - 4:
Western blot - Human RIPK1 knockout THP-1 cell line (ab276121)

Lane 1:
Wild-type THP-1 cell lysate at 20 µg

Lane 2:
RIPK1 knockout THP-1 cell lysate at 20 µg

Lane 3:
HeLa cell lysate at 20 µg

Lane 4:
Raji cell lysate at 20 µg

Developed using the ECL technique.

Performed under reducing conditions.

Predicted band size: 75 kDa

Observed band size: 76 kDa

Please note: All products are 'FOR RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC OR THERAPEUTIC PROCEDURES'.